

DETERMINATION OF INORGANIC ANIONS BY ION CHROMATOGRAPHY

1.0 SCOPE AND APPLICATION

1.1 This method addresses the sequential determination of the anions chloride, fluoride, bromide, nitrate, nitrite, phosphate, and sulfate in the collection solutions from the bomb combustion of solid waste samples, as well as all water samples.

1.2 The method detection limit (MDL), the minimum concentration of a substance that can be measured and reported with 99% confidence that the value is above zero, varies for anions as a function of sample size and the conductivity scale used. Generally, minimum detectable concentrations are in the range of 0.05 mg/L for F^- and 0.1 mg/L for Br^- , Cl^- , NO_3^- , NO_2^- , PO_4^{3-} , and SO_4^{2-} with a 100- μ L sample loop and a 10- μ mho full-scale setting on the conductivity detector. Similar values may be achieved by using a higher scale setting and an electronic integrator. Idealized detection limits of an order of magnitude lower have been determined in reagent water by using a 1- μ mho/cm full-scale setting (Table 1). The upper limit of the method is dependent on total anion concentration and may be determined experimentally. These limits may be extended by appropriate dilution.

2.0 SUMMARY OF METHOD

2.1 A small volume of combustate collection solution or other water sample, typically 2 to 3 mL, is injected into an ion chromatograph to flush and fill a constant volume sample loop. The sample is then injected into a stream of carbonate-bicarbonate eluent of the same strength as the collection solution or water sample.

2.2 The sample is pumped through three different ion exchange columns and into a conductivity detector. The first two columns, a precolumn or guard column and a separator column, are packed with low-capacity, strongly basic anion exchanger. Ions are separated into discrete bands based on their affinity for the exchange sites of the resin. The last column is a suppressor column that reduces the background conductivity of the eluent to a low or negligible level and converts the anions in the sample to their corresponding acids. The separated anions in their acid form are measured using an electrical-conductivity cell. Anions are identified based on their retention times compared to known standards. Quantitation is accomplished by measuring the peak height or area and comparing it to a calibration curve generated from known standards.

3.0 INTERFERENCES

3.1 Any species with a retention time similar to that of the desired ion will interfere. Large quantities of ions eluting close to the ion of interest will also result in an interference. Separation can be improved by adjusting the eluent concentration and/or flow rate. Sample dilution and/or the use of the

method of standard additions can also be used. For example, high levels of organic acids may be present in industrial wastes, which may interfere with inorganic anion analysis. Two common species, formate and acetate, elute between fluoride and chloride.

3.2 Because bromide and nitrate elute very close together, they are potential interferences for each other. It is advisable not to have $\text{Br}^-/\text{NO}_3^-$ ratios higher than 1:10 or 10:1 if both anions are to be quantified. If nitrate is observed to be an interference with bromide, use of an alternate detector (e.g., electrochemical detector) is recommended.

3.3 Method interferences may be caused by contaminants in the reagent water, reagents, glassware, and other sample processing apparatus that lead to discrete artifacts or elevated baseline in ion chromatograms.

3.4 Samples that contain particles larger than 0.45 μm and reagent solutions that contain particles larger than 0.20 μm require filtration to prevent damage to instrument columns and flow systems.

3.5 If a packed bed suppressor column is used, it will be slowly consumed during analysis and, therefore, will need to be regenerated. Use of either an anion fiber suppressor or an anion micromembrane suppressor eliminates the time-consuming regeneration step through the use of a continuous flow of regenerant.

4.0 APPARATUS AND MATERIALS

4.1 Ion chromatograph, capable of delivering 2 to 5 mL of eluent per minute at a pressure of 200 to 700 psi (1.3 to 4.8 MPa). The chromatograph shall be equipped with an injection valve, a 100- μL sample loop, and set up with the following components, as schematically illustrated in Figure 1.

4.1.1 Precolumn, a guard column placed before the separator column to protect the separator column from being fouled by particulates or certain organic constituents (4 x 50 mm, Dionex P/N 030825 [normal run], or P/N 030830 [fast run], or equivalent).

4.1.2 Separator column, a column packed with low-capacity pellicular anion exchange resin that is styrene divinylbenzene-based has been found to be suitable for resolving F^- , Cl^- , NO_2^- , PO_4^{3-} , Br^- , NO_3^- , and SO_4^{2-} (see Figure 2) (4 x 250 mm, Dionex P/N 03827 [normal run], or P/N 030831 [fast run], or equivalent).

4.1.3 Suppressor column, a column that is capable of converting the eluent and separated anions to their respective acid forms (fiber, Dionex P/N 35350, micromembrane, Dionex P/N 38019 or equivalent).

4.1.4 Detector, a low-volume, flowthrough, temperature-compensated, electrical conductivity cell (approximately 6 μL volume, Dionex, or equivalent) equipped with a meter capable of reading from 0 to 1,000 $\mu\text{seconds/cm}$ on a linear scale.

4.1.5 Pump, capable of delivering a constant flow of approximately 2 to 5 mL/min throughout the test and tolerating a pressure of 200 to 700 psi (1.3 to 4.8 MPa).

4.2 Recorder, compatible with the detector output with a full-scale response time in 2 seconds or less.

4.3 Syringe, minimum capacity of 2 mL and equipped with a male pressure fitting.

4.4 Eluent and regenerant reservoirs, suitable containers for storing eluents and regenerant. For example, 4 L collapsible bags can be used.

4.5 Integrator, to integrate the area under the chromatogram. Different integrators can perform this task when compatible with the electronics of the detector meter or recorder. If an integrator is used, the maximum area measurement must be within the linear range of the integrator.

4.6 Analytical balance, capable of weighing to the nearest 0.0001 g.

4.7 Pipets, Class A volumetric flasks, beakers: assorted sizes.

5.0 REAGENTS

5.1 Reagent grade chemicals shall be used in all tests. Unless otherwise indicated, it is intended that all reagents shall conform to the specifications of the Committee on Analytical Reagents of the American Chemical Society, where such specifications are available. Other grades may be used, provided it is first ascertained that the reagent is of sufficiently high purity to permit its use without lessening the accuracy of the determination.

5.2 Reagent water. All references to water in this method refer to reagent water, as defined in Chapter One. Column life may be extended by passing reagent water through a 0.22- μ m filter prior to use.

5.3 Eluent, 0.003M NaHCO₃/0.0024M Na₂CO₃. Dissolve 1.0080 g of sodium bicarbonate (0.003M NaHCO₃) and 1.0176 g of sodium carbonate (0.0024M Na₂CO₃) in reagent water and dilute to 4 L with reagent water.

5.4 Suppressor regenerant solution. Add 100 mL of 1N H₂SO₄ to 3 L of reagent water in a collapsible bag and dilute to 4 L with reagent water.

5.5 Stock solutions (1,000 mg/L).

5.5.1 Bromide stock solution (1.00 mL = 1.00 mg Br⁻). Dry approximately 2 g of sodium bromide (NaBr) for 6 hours at 150°C, and cool in a desiccator. Dissolve 1.2877 g of the dried salt in reagent water, and dilute to 1 L with reagent water.

5.5.2 Chloride stock solution (1.00 mL = 1.00 mg Cl⁻). Dry sodium chloride (NaCl) for 1 hour at 600°C, and cool in a desiccator. Dissolve 1.6484 g of the dry salt in reagent water, and dilute to 1 L with reagent water.

5.5.3 Fluoride stock solution (1.00 mL = 1.00 mg F⁻). Dissolve 2.2100 g of sodium fluoride (NaF) in reagent water, and dilute to 1 L with reagent water. Store in chemical-resistant glass or polyethylene.

5.5.4 Nitrate stock solution (1.00 mL = 1.00 mg NO_3^-). Dry approximately 2 g of sodium nitrate (NaNO_3) at 105°C for 24 hours. Dissolve exactly 1.3707 g of the dried salt in reagent water, and dilute to 1 L with reagent water.

5.5.5 Nitrite stock solution (1.00 mL = 1.00 mg NO_2^-). Place approximately 2 g of sodium nitrate (NaNO_2) in a 125 mL beaker and dry to constant weight (about 24 hours) in a desiccator containing concentrated H_2SO_4 . Dissolve 1.4998 g of the dried salt in reagent water, and dilute to 1 L with reagent water. Store in a sterilized glass bottle. Refrigerate and prepare monthly.

NOTE: Nitrite is easily oxidized, especially in the presence of moisture, and only fresh reagents are to be used.

NOTE: Prepare sterile bottles for storing nitrite solutions by heating for 1 hour at 170°C in an air oven.

5.5.6 Phosphate stock solution (1.00 mL = 1.00 mg PO_4^{3-}). Dissolve 1.4330 g of potassium dihydrogen phosphate (KH_2PO_4) in reagent water, and dilute to 1 L with reagent water. Dry sodium sulfate (Na_2SO_4) for 1 hour at 105°C and cool in a desiccator.

5.5.7 Sulfate stock solution (1.00 mL = 1.00 mg SO_4^{2-}). Dissolve 1.4790 g of the dried salt in reagent water, and dilute to 1 L with reagent water.

5.6 Anion working solutions. Prepare a blank and at least three different working solutions containing the following combinations of anions. The combination anion solutions must be prepared in Class A volumetric flasks. See Table 2.

5.6.1 Prepare a high-range standard solution by diluting the volumes of each anion specified in Table 2 together to 1 L with reagent water.

5.6.2 Prepare the intermediate-range standard solution by diluting 10.0 mL of the high-range standard solution (see Table 2) to 100 mL with reagent water.

5.6.3 Prepare the low-range standard solution by diluting 20.0 mL of the intermediate-range standard solution (see Table 2) to 100 mL with reagent water.

5.7 Stability of standards. Stock standards are stable for at least 1 month when stored at 4°C. Dilute working standards should be prepared weekly, except those that contain nitrite and phosphate, which should be prepared fresh daily.

6.0 SAMPLE COLLECTION, PRESERVATION, AND HANDLING

6.1 All samples must have been collected using a sampling plan that addresses the considerations discussed in Chapter Nine of this manual.

6.2 Analyze the samples as soon as possible after collection. Preserve by refrigeration at 4°C.

7.0 PROCEDURE

7.1 Calibration

7.1.1 Establish ion chromatographic operating parameters equivalent to those indicated in Table 1.

7.1.2 For each analyte of interest, prepare calibration standards at a minimum of three concentration levels and a blank by adding accurately measured volumes of one or more stock standards to a Class A volumetric flask and diluting to volume with reagent water. If the working range exceeds the linear range of the system, a sufficient number of standards must be analyzed to allow an accurate calibration curve to be established. One of the standards should be representative of a concentration near, but above, the method detection limit if the system is operated on an applicable attenuator range. The other standards should correspond to the range of concentrations expected in the sample or should define the working range of the detector. Unless the attenuator range settings are proven to be linear, each setting must be calibrated individually.

7.1.3 Using injections of 0.1 to 1.0 mL (determined by injection loop volume) of each calibration standard, tabulate peak height or area responses against the concentration. The results are used to prepare a calibration curve for each analyte. During this procedure, retention times must be recorded.

7.1.4 The working calibration curve must be verified on each working day, or whenever the anion eluent strength is changed, and for every batch of samples. If the response or retention time for any analyte varies from the expected values by more than $\pm 10\%$, the test must be repeated, using fresh calibration standards. If the results are still more than $\pm 10\%$, an entirely new calibration curve must be prepared for that analyte.

7.1.5 Nonlinear response can result when the separator column capacity is exceeded (overloading). Maximum column loading (all anions) should not exceed about 400 ppm.

7.2 Analyses

7.2.1 Sample preparation. When aqueous samples are injected, the water passes rapidly through the columns, and a negative "water dip" is observed that may interfere with the early-eluting fluoride and/or chloride ions. The water dip should not be observed in the combustate samples; the collecting solution is a concentrated eluent solution that will "match" the eluent strength when diluted to 100-mL with reagent water according to the bomb combustion procedure. Any dilutions required in analyzing other water samples should be made with the eluent solution. The water dip, if present, may be removed by adding concentrated eluent to

all samples and standards. When a manual system is used, it is necessary to micropipet concentrated buffer into each sample. The recommended procedures follow:

- (1) Prepare a 100-mL stock of eluent 100 times normal concentration by dissolving 2.5202 g NaHCO_3 and 2.5438 g Na_2CO_3 in 100-mL reagent water. Protect the volumetric flask from air.
- (2) Pipet 5 mL of each sample into a clean polystyrene micro-beaker. Micropipet 50 μL of the concentrated buffer into the beaker and stir well.

Dilute the samples with eluent, if necessary, to concentrations within the linear range of the calibration.

7.2.2 Sample analysis.

7.2.2.1 Start the flow of regenerant through the suppressor column.

7.2.2.2 Set up the recorder range for maximum sensitivity and any additional ranges needed.

7.2.2.3 Begin to pump the eluent through the columns. After a stable baseline is obtained, inject a midrange standard. If the peak height deviates by more than 10% from that of the previous run, prepare fresh standards.

7.2.2.4 Begin to inject standards starting with the highest concentration standard and decreasing in concentration. The first sample should be a quality control reference sample to check the calibration.

7.2.2.5 Using the procedures described in Step 7.2.1, calculate the regression parameters for the initial standard curve. Compare these values with those obtained in the past. If they exceed the control limits, stop the analysis and look for the problem.

7.2.2.6 Inject a quality control reference sample. A spiked sample or a sample of known content must be analyzed with each batch of samples. Calculate the concentration from the calibration curve and compare the known value. If the control limits are exceeded, stop the analysis until the problem is found. Recalibration is necessary.

7.2.2.7 When an acceptable value has been obtained for the quality control sample, begin to inject the samples.

7.2.2.8 Load and inject a fixed amount of well-mixed sample. Flush injection loop thoroughly, using each new sample.

Use the same size loop for standards and samples. Record the resulting peak size in area or peak height units. An automated constant volume injection system may also be used.

7.2.2.9 The width of the retention time window used to make identifications should be based on measurements of actual retention time variations of standards over the course of a day. Three times the standard deviation of a retention time can be used to calculate a suggested window size for a compound. However, the experience of the analyst should weigh heavily in the interpretation of chromatograms.

7.2.2.10 If the response for the peak exceeds the working range of the system, dilute the sample with an appropriate amount of reagent water and reanalyze.

7.2.2.11 If the resulting chromatogram fails to produce adequate resolution, or if identification of specific anions is questionable, spike the sample with an appropriate amount of standard and reanalyze.

NOTE: Nitrate and sulfate exhibit the greatest amount of change, although all anions are affected to some degree. In some cases, this peak migration can produce poor resolution or misidentification.

7.3 Calculation

7.3.1 Prepare separate calibration curves for each anion of interest by plotting peak size in area, or peak height units of standards against concentration values. Compute sample concentration by comparing sample peak response with the standard curve.

7.3.2 Enter the calibration standard concentrations and peak heights from the integrator or recorder into a calculator with linear least squares capabilities.

7.3.3 Calculate the following parameters: slope (s), intercept (I), and correlation coefficient (r). The slope and intercept define a relationship between the concentration and instrument response of the form:

$$y_i = s_i x_i + I \quad (1)$$

where:

y_i = predicted instrument response
 s_i = response slope
 x_i = concentration of standard i
I = intercept

Rearrangement of the above equation yields the concentration corresponding to an instrumental measurement:

$$x_j = (y_j - I)/s_j \quad (2)$$

where:

x_j = calculated concentration for a sample
 y_j = actual instrument response for a sample
 s_j and I are calculated slope and intercept from calibration above.

7.3.4 Enter the sample peak height into the calculator, and calculate the sample concentration in milligrams per liter.

8.0 QUALITY CONTROL

8.1 All quality control data should be maintained and available for easy reference and inspection. Refer to Chapter One for additional quality control guidelines.

8.2 After every 10 injections, analyze a midrange calibration standard. If the instrument response has changed by more than 5%, recalibrate.

8.3 Analyze one in every ten samples in duplicate. Take the duplicate sample through the entire sample preparation and analytical process.

8.4 A matrix spiked sample should be run for each analytical batch or twenty samples, whatever is more frequent, to determine matrix effects.

9.0 METHOD PERFORMANCE

9.1 Single-operator accuracy and precision for reagent, drinking and surface water, and mixed domestic and industrial wastewater are listed in Table 3.

9.2 Combustate samples. These data are based on 41 data points obtained by six laboratories who each analyzed four used crankcase oils and three fuel oil blends with crankcase in duplicate. The oil samples were combusted using Method 5050. A data point represents one duplicate analysis of a sample. One data point was judged to be an outlier and was not included in the results.

9.2.1 Precision. The precision of the method as determined by the statistical examination of interlaboratory test results is as follows:

Repeatability - The difference between successive results obtained by the sample operator with the same apparatus under constant operating conditions on identical test material would exceed, in the long run, in the normal and correct operation of the test method, the following values only in 1 case in 20 (see Table 4):

*where x is the average of two results in $\mu\text{g/g}$.

$$\text{Repeatability} = 20.9 \sqrt{x}^*$$

Reproducibility - The difference between two single and independent results obtained by different operators working in different laboratories on identical test material would exceed, in the long run, the following values only in 1 case in 20:

$$\text{Reproducibility} = 42.1 \sqrt{x}^*$$

*where x is the average value of two results in µg/g.

9.2.2 Bias. The bias of this method varies with concentration, as shown in Table 5:

$$\text{Bias} = \text{Amount found} - \text{Amount expected}$$

10.0 REFERENCES

1. Environmental Protection Agency. Test Method for the Determination of Inorganic Anions in Water by Ion Chromatography. EPA Method 300.0. EPA-600/4-84-017. 1984.
2. Annual Book of ASTM Standards, Volume 11.01 Water D4327, Standard Test Method for Anions in Water by Ion Chromatography, pp. 696-703. 1988.
3. Standard Methods for the Examination of Water and Wastewater, Method 429, "Determination of Anions by Ion Chromatography with Conductivity Measurement," 16th Edition of Standard Methods.
4. Dionex, IC 16 Operation and Maintenance Manual, PN 30579, Dionex Corp., Sunnyvale, CA 94086.
5. Method detection limit (MDL) as described in "Trace Analyses for Wastewater," J. Glaser, D. Foerst, G. McKee, S. Quave, W. Budde, Environmental Science and Technology, Vol. 15, Number 12, p. 1426, December 1981.
6. Gaskill, A.; Estes, E. D.; Hardison, D. L.; and Myers, L. E. Validation of Methods for Determining Chlorine in Used Oils and Oil Fuels. Prepared for U.S. Environmental Protection Agency Office of Solid Waste. EPA Contract No. 68-01-7075, WA 80. July 1988.

TABLE 1.
CHROMATOGRAPHIC CONDITIONS AND METHOD DETECTION
LIMITS IN REAGENT WATER

Analyte	Retention ^a time min	Relative retention time	Method ^b detection limit, mg/L
Fluoride	1.2	1.0	0.005
Chlorine	3.4	2.8	0.015
Nitrite-N	4.5	3.8	0.004
o-Phosphate-P	9.0	7.5	0.061
Nitrate-N	11.3	9.4	0.013
Sulfate	21.4	17.8	0.206

Standard conditions:

Columns - As specified in 4.1.1-4.1.3
 Detector - As specified in 4.1.4
 Eluent - As specified in 5.3

Sample loop - 100 µL
 Pump volume - 2.30 mL/min

Concentrations of mixed standard (mg/L):

Fluoride 3.0
 Chloride 4.0
 Nitrite-N 10.0

o-Phosphate-P 9.0
 Nitrate-N 30.0
 Sulfate 50.0

^aThe retention time given for each anion is based on the equipment and analytical conditions described in the method. Use of other analytical columns or different elutant concentrations will effect retention times accordingly.

^bMDL calculated from data obtained using an attenuator setting of 1-µmho/cm full scale. Other settings would produce an MDL proportional to their value.

TABLE 2.
PREPARATION OF STANDARD SOLUTIONS FOR INSTRUMENT CALIBRATION

	High Range Standard ¹	Anion concentration mg/L	Intermediate- range standard, mg/L (see 5.6.2)	Low-range standard, mg/L (see 5.6.3)
Fluoride (F ⁻)	10	10	1.0	0.2
Chloride (Cl ⁻)	10	10	1.0	0.2
Nitrite (NO ₂ ⁻)	20	20	2.0	0.4
Phosphate (PO ₄ ³⁻)	50	50	5.0	1.0
Bromide (Br ⁻)	10	10	1.0	0.2
Nitrate (NO ₃ ⁻)	30	30	3.0	0.6
Sulfate (SO ₄ ²⁻)	100	100	10.0	2.0

¹Milliliters of each stock solution (1.00 mL = 1.00 mg) diluted to 1 L (see sec. 5.6.1).

TABLE 3.
SINGLE-OPERATOR ACCURACY AND PRECISION

Analyte	Sample type	Spike mg/L	Number of replicates	Mean recovery, %	Standard deviation, mg/L
Chloride	RW	0.050	7	97.7	0.0047
	DW	10.0	7	98.2	0.289
	SW	1.0	7	105.0	0.139
	WW	7.5	7	82.7	0.445
Fluoride	RW	0.24	7	103.1	0.0009
	DW	9.3	7	87.7	0.075
	SW	0.50	7	74.0	0.0038
	WW	1.0	7	92.0	0.011
Nitrate-N	RW	0.10	7	100.9	0.0041
	DW	31.0	7	100.7	0.356
	SW	0.50	7	100.0	0.0058
	WW	4.0	7	94.3	0.058
Nitrite-N	RW	0.10	7	97.7	0.0014
	DW	19.6	7	103.3	0.150
	SW	0.51	7	88.2	0.0053
	WW	0.52	7	100.0	0.018
o-Phosphate-P	RW	0.50	7	100.4	0.019
	DE	45.7	7	102.5	0.386
	SW	0.51	7	94.1	0.020
	WW	4.0	7	97.3	0.04
Sulfate	RW	1.02	7	102.1	0.066
	DW	98.5	7	104.3	1.475
	SW	10.0	7	111.6	0.709
	WW	12.5	7	134.9	0.466

RW = Reagent water.
DW = Drinking water.

SW = Surface water.
WW = Wastewater.

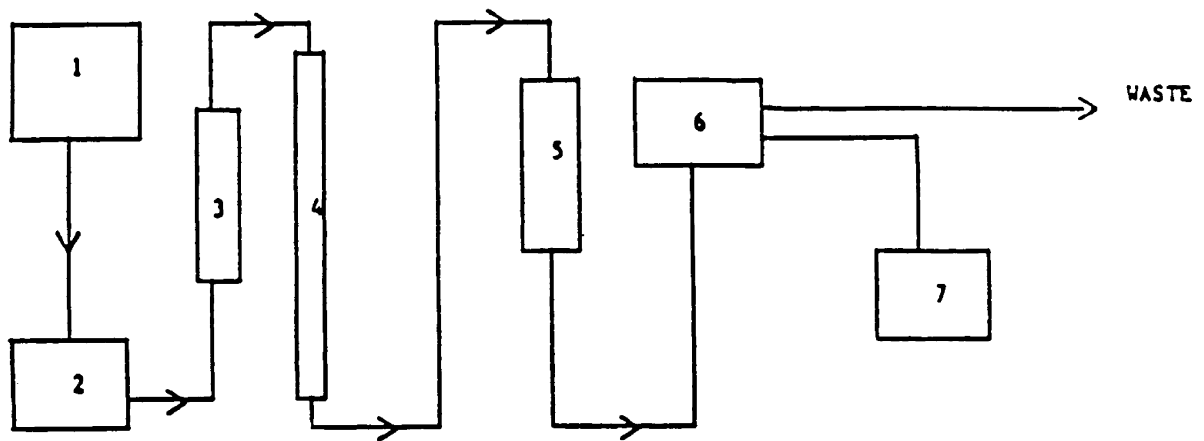
TABLE 4.
REPEATABILITY AND REPRODUCIBILITY FOR CHLORINE IN
USED OILS BY BOMB OXIDATION AND ION CHROMATOGRAPHY

Average value, $\mu\text{g/g}$	Repeatability, $\mu\text{g/g}$	Reproducibility, $\mu\text{g/g}$
500	467	941
1,000	661	1,331
1,500	809	1,631
2,000	935	1,883
2,500	1,045	2,105
3,000	1,145	2,306

TABLE 5.
RECOVERY AND BIAS DATA FOR CHLORINE IN USED OILS BY
BOMB OXIDATION AND ION CHROMATOGRAPHY

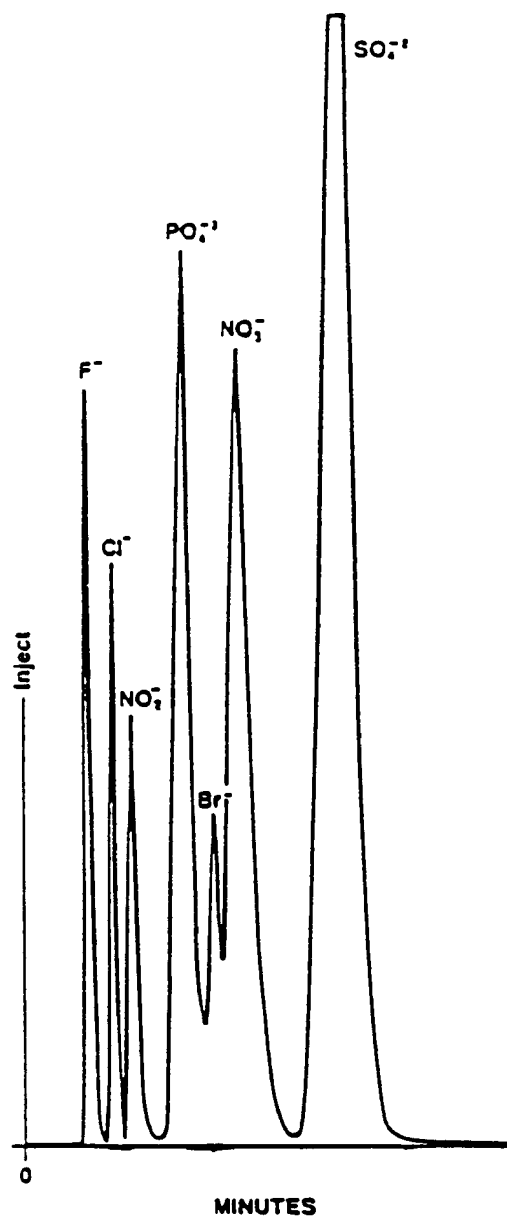
Amount Expected $\mu\text{g/g}$	Amount found $\mu\text{g/g}$	Bias, $\mu\text{g/g}$	Percent, bias
320	567	247	+77
480	773	293	+61
920	1,050	130	+14
1,498	1,694	196	+13
1,527	1,772	245	+16
3,029	3,026	-3	0
3,045	2,745	-300	-10

FIGURE 1
SCHEMATIC OF ION CHROMATOGRAPH



- (1) Eluent reservoir
- (2) Pump
- (3) Precolumn
- (4) Separator column
- (5) Suppressor column
- (6) Detector
- (7) Recorder or integrator, or both

FIGURE 2
TYPICAL ANION PROFILE



METHOD 9056
DETERMINATION OF INORGANIC ANIONS BY ION CHROMATOGRAPHY

